and microtubules. Since APC is known to be truncated in many cancer cells, the authors analyzed a glioblastoma cell line where APC was barely detectable and found that the intermediate filament network was retracted towards the nucleus. This observation is not true in all cancer cells, however, as it turns out that APC does not interact with keratin intermediate filaments, and the keratin network remains well spread in the absence of APC, even though the vimentin network is retracted.

The authors also found that the region of APC that interacted with intermediate filaments was located in the armadillo repeat motif (ARM) closer to the amino terminus of APC. Thus, the domains that interact with intermediate filaments and microtubules are well separated in the molecule. Immunoprecipitation experiments confirmed that both vimentin and GFAP interact with the ARM region. *In vitro* polymerization assays suggest that this region of APC enhances intermediate filament polymerization, at least as measured by turbidity. Since no EM data were presented for these experiments, it is hard to determine whether the ARM region of APC caused elongation of intermediate filaments, although the intensity of immunofluorescence staining increased when vimentin was polymerized in the presence of the amino-terminal region of APC.

What is the significance of these interactions between microtubules and intermediate filaments? As is well-known, microtubules are important for cell polarization and migration, as well as for centrosome reorientation. To study the importance of the APC-mediated connection between the two cytoskeletal elements, the authors microinjected astrocytes migrating towards a wound edge with a number of APC constructs, including two that contain the intermediate-filament-binding site and one that did not. The results showed that the constructs that contained the intermediate-filament-binding site resulted in a collapse of the intermediate filaments and inhibited astrocyte migration, but had no effect on centrosome reorientation. As further evidence that intermediate filaments are involved in migration, the authors also used a carboxy-terminal dominant-negative GFAP construct, which collapses the intermediate filament network, and found that this construct also inhibited migration.

The studies in this paper therefore reveal that the tumor suppressor APC is able to interact with microtubules and intermediate filaments through two distinct regions and that APC interacts with two types of intermediate filaments, i.e. GFAP and vimentin, although not with keratins. It remains to be seen whether neuronal intermediate filaments also interact with microtubules through APC, or whether these interactions are primarily through microtubule motor proteins, which function to transport the neuronal intermediate filaments along the axon.

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Invertebrate Neurobiology: Short-Term Memories for Limb Targeting

Scanning movements made by stick insects' forelimbs are modified for several seconds after a brief contact with an object, suggesting that the neural networks controlling local limb movements in insects can form short-term positional memories.

Jeremy E. Niven

Walking across a darkened room your vision is of little use. Instead, you're likely to make searching movements with your arms through the space ahead of you to detect objects in your path. Most of the space will probably be empty but when you encounter an object, your movements will change as you try to identify it; no longer sweeping through the space, but making small directed movements towards the object. Without vision your progress across the room will be slow.

For humans, this strategy is a backup, implemented when vision is poor. For many walking insects, searching the space ahead with forelimbs or antennae is essential for detecting and locating obstacles or footholds, and targeting their limbs towards them [1,2]. The searching movements of most insects are typically rhythmic, sweeping through a large region of space, but what happens to these movements once they encounter an object? In a recent paper, Berg *et al.* [3] show that,

once a stick insect's forelimb makes contact with an object, the forelimbs movements are modified for several seconds by a short-term memory so that it targets the position of the object.

Berg *et al.* [3] used a preparation in which a stick insect (*Cuniculina impigra*) could move only its left forelimb. They evoked rhythmic searching movements of the forelimb by touching or puffing air at the stick insect. After four cycles of uninterrupted searching movements, a stick was inserted into the forelimb's plane of movement, so that the distal tibia of forelimb would make contact with it. The stick was inserted at one of four different positions, and was retracted immediately after contact so that there was only a single point of contact between the forelimb and the stick. Following contact, the researchers observed that the forelimb's rhythmic searching movements were modified so that their average position was shifted towards the contact point and their amplitude dropped. By inserting the stick at different positions, the authors could ensure that the change they observed in the searching movements was not a stereotyped response but rather a specific response to the contact.

Kinematic analysis showed that both modifications of the forelimb's searching movements were due primarily to changes in the coxa-trochanteral joint of the leg. Berg *et al.* [3] then sought to identify the sense organs necessary for targeting the forelimb's movements to the stick's position. They removed the distal portion of the leg and replaced it with a wooden stick to prevent distal sense organs, especially those on the tibia, from signalling contact with the object; however, this 'peg leg' did not affect the stick insects' ability to perform a targeted response in the majority of experiments. Next, Berg *et al.* [3] ablated a proximal leg mechanosensory organ, the trochanteral hair plate, which has been implicated in control of the coxa-trochanteral joint [4]. Trochanteral hair plate ablation reduced the number of experiments in which the position and amplitude was modified. Ablation did not, however, prevent modification of the forelimb movements entirely, consistent with the presence of numerous sense organs operating in parallel in the insect leg [5].

The modification of the forelimb searching movements persisted for several seconds after contact was made [3]. This modification is likely to be generated actively because it depends upon which of the four possible positions was the contact point. This implies a memory is formed of the forelimbs' position at the contact point. The memory is short-term, lasting only around six seconds before the forelimbs' movements return to the same position and amplitude as before contact. This is reminiscent of the short-term memory proposed to account for persistent forelimb targeting in locusts in a ladder-walking paradigm [6]. The locusts continue to target their forelimbs to the location of a particular ladder rung even after the rung has been shifted to a new location for several seconds before re-targeting to the new rung position. Unlike the stick insects, however, the locusts use visual cues to target their forelimbs to the rung. A similar short-term memory may also account for the persistent hind limb scratching movements in locusts [6]. These scratching movements can be evoked by a brief touch of the locusts' wings with a paint brush. The rhythmic scratching movements elicited are targeted to the contact point and can continue for many cycles after the cessation of the stimulus [7].

The experiments of Berg *et al.* [3] imply that the local circuits that control limb movements in stick insects are capable of storing short-term memories of specific positions. These are also the circuits in which long-term memories of leg position can be evoked with aversive conditioning in cockroaches and locusts [8]. As Berg *et al.* [3] point out, however, the short-term memories for limb targeting are quite different from long-term memories of leg position in locusts and cockroaches, not only in their duration but also in the likely mechanisms involved. These local limb control networks may also be the site of the short-term memory of rung position in the ladder walking locusts and of the tactile stimulus position on the wing that evokes scratching in locusts, though additional experiments will be needed to test this.

Numerous mechanisms could account for the short-term memory of position in the local limb control networks in insects. Berg *et al.* [3] suggest several potential mechanisms based on the intrinsic membrane properties of neurons that could produce a cellular memory trace. For example, voltage-gated K^+ channels can have slow kinetics that can act as a short-term cellular memory [9]. Another possibility, however, is short-term plasticity at the synapses between neurons in the local networks. Short-term depression has been shown between the synapses of exteroceptors from hairs on the hind leg of the locust and spiking local interneurons that form part of the local networks controlling movements of the hind legs [10]. Moreover, computational modelling has shown that short-term synaptic plasticity could act as a mechanism for the maintenance of working memory [11] and can operate over the necessary timescales to account for the short-term memory of contact position in stick insects and locusts.

The local limb control networks of locusts and stick insects have been described in considerable detail and the numerous sense organs, populations of interneurons and motor neurons have been identified [5]. Thus, Berg *et al.* [3] have developed a paradigm in which it should be possible to identify the mechanisms underpinning this short-term memory in these local networks of spiking and non-spiking neurons and relate these mechanisms directly to the movements of the forelimb. This has the potential to reveal how an intact neural network maintains such short-term memories *in vivo*.

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Diet and Genetics: Trp-ing Over Food Sensitivity

Laboratory-reared *Caenorhabditis elegans* eat *Escherichia coli*. A new study demonstrates a strong diet–gene interaction: worms with reduced *nhr-114* activity are fertile when fed *E. coli* K-12 strains but are sterile on *E. coli* B. Surprisingly, tryptophan supplementation of *E. coli* B restores worm fertility.

E. Jane Albert Hubbard

"You are what you eat", "one man's meat is another man's poison", "nature versus nurture": such adages reflect a long-standing appreciation of the influence of diet on health, different responses to diet between individuals, and the roles of both diet and genetics in shaping phenotype. The additional connection between diet and fertility is an ever-expanding area of interest both in the scientific and popular literature. A recent web search of "fertility diet" returns over 150,000 hits. How do genetic differences influence the response to food? And how do diet and genetics conspire to influence fertility?

Using *Caenorhabditis elegans* as a model, a recent study published in *Current Biology* by Gracida and Eckmann [1] demonstrates how a combination of diet and genetics can radically influence fertility. The key finding is that the activity of a *C. elegans* HNF4-like nuclear hormone receptor (NHR), *nhr-114*, is the critical arbiter of fertility versus sterility, but only in combination with certain *Escherichia coli* food sources. Specifically, *nhr-114*-deficient worms are fertile when fed on strains derived from *E. coli* K-12 but are sterile when fed on a diet of *E. coli* B-derived strains (Figure 1). The germline defects underlying sterility in animals with reduced *nhr-114* activity on a diet of *E. coli* B strains include a failure to produce oocytes, defects in cell division and overall loss of nuclear and cellular integrity of the proliferating pool of germline stem cells. These defects first appear during larval development when the germline stem cell pool would normally expand. In addition, most of the germline defects were rescued by *nhr-114* activity in the intestine.

NHR-114 is one of over 250 NHRs encoded in the *C. elegans* genome, most of which are thought to have arisen by duplication from a common HNF4-related ancestor [2]. Other so-called 'supplementary nuclear receptors' have been implicated in many aspects of *C. elegans* biology, including metabolism, morphogenesis, longevity and the germline [3–6].

Remarkably, the authors determined that the sterility caused by the combination of *nhr-114* depletion and an *E. coli* B diet could be circumvented by prior supplementation of the live bacteria with tryptophan (Trp). That is, *nhr-114*-deficient worms fed on *E. coli* B supplemented with Trp are fertile (Figure 1). While Trp is an essential amino acid for *C. elegans* [7], the effect is not likely due to direct uptake of Trp by the worms since *nhr-114*-depleted worms fed heat- or UV-killed *E. coli* B bacteria supplemented with Trp were not fertile. However, *nhr-114* worms fed dead bacteria were fertile, provided the bacteria were supplemented with Trp for a sufficient time while they were still alive.

Transcriptional profile analysis suggested an underlying role for detoxification pathways as a plausible mechanistic basis for the observed differences in the phenotypic responses to diet and *nhr-114*. The authors compared global patterns of gene expression of wild-type worms fed the *E. coli* B-derived OP50 strain with and without Trp supplementation and found that genes associated with detoxification and xenobiotic responses were induced by Trp supplementation. Next, they identified genes whose expression was altered by depletion of *nhr-114* compared with a strain that essentially lacks germ cells. Among the ~2000 genes in the latter set, over a quarter overlapped with the Trp set. Of these, over half were reciprocally regulated (down in *nhr-114(–)* and up in Trp supplementation), as would be expected if Trp supplementation and *nhr-114(+)* activity were acting similarly. Notably, this set includes genes regulated by HNF4 in mammals.

The model proposed from these studies is that scarce Trp or reduced Trp metabolism in the *E. coli* B-derived strain leads to the production of toxins or toxic metabolites that are normally cleared by *nhr-114*-dependent processes. When *nhr-114* activity is reduced in the intestine, the germline becomes susceptible to the adverse effects of these toxic dietary metabolites, leading to progressive germline failure and sterility. In this model, *nhr-114(+)* acts as a buffer to preserve fertility in the presence of toxic metabolites introduced by certain diets. A related model is that *nhr-114*(+) promotes germline maintenance in parallel with a Trp-dosage-dependent metabolite, such that elevating the concentration of Trp compensates for loss of *nhr-114*. In this model, the *E. coli* K-12 strain would presumably supply the necessary factor even in low Trp conditions. Regardless, Trp supplementation alters the metabolic landscape of *E. coli* B-derived bacteria such that *nhr-114* is no longer necessary for worm fertility. The key differences between *E. coli* B- and K12-derived strains that underlie the exquisite sensitivity to Trp (as far as their utility as worm food is concerned), as well as the nature of the putative toxic substances